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DURATION OF IMMATURE STAGES OF ELEVEN SWALLOWTAIL BUTTERFLIES (LEPIDOPTERA: PAPILIONIDAE) IN WEST SUMATRA, INDONESIA

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The duration of the immature stages of 11 Sumatran swallowtail butterflies was examined under laboratory conditions. The total developmental times of seven Papilionini species (average 43.6 days) and one Graphiini species (39.8 days) were shorter than those of Troidini species (62.1 days). Among these 11 species, *Papilio nephelus* showed the largest variation in the duration of its immature stages (35-141 days) due to a variable pupal stage and the longest total developmental time (average 78 days). Fifty six percent of *P. nephelus* pupae were considered to enter diapause. All individuals of *Pachliopta aristolochiae* passed through six instars while the other 10 species had five. We compared the developmental times of the present species with those of related species reported previously and discuss the variation in larval period in relation to diapause and the extra instar.

KEY WORDS: Lepidoptera, Papilionidae, duration of immature stages, *Papilio, Graphium*, *Pachliopta, Troides, Trogonoptera,* extra instar, diapause, West Sumatra, Indonesia.

Дахелми[1], С. Салмах[2], И. Аббас[2], Н. Фитрайна[2], С. Накано[3], К. Накамура[1,4]. Продолжительность предимагинальных стадий одиннадцати видов хвостоносцев (Lepidoptera: Papilionidae) на Восточной Суматре, Индонезия // Дальневосточный энтомолог. 2008. N 182. С. 1-9.

В лабораторных условиях изучена продолжительность предимагинального развития 11 видов суматранских хвостоносцев. Показано, что период развития у 7 видов трибы Papilionini (в среднем 43.6 дней) и 1 вида Graphiini (39.8 дней) ясно короче, чем у 3 видов трибы Troidini (в среднем 62.1 дней). Из 11 изученных видов наибольшая вариабельность длительности предимагинального развития отмечена у *Papilio nephelus* (35-141 дней), что связано с продолжительностью стадии куколки и наибольшей длительностью всего развития (около 78 дней). В диапаузу впадает 56% куколок *P. nephelus.* Количество возрастов гусениц у изученных видов – пять, только у *Pachliopta aristolochiae* – шесть. Проведено сравнение времени развития у исследованных видов с таковым у изученных ранее близких видов и обсуждена продолжительность развития в зависимости от диапаузы и наличия дополнительного возраста у гусениц.

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INTRODUCTION

Swallowtail butterflies (family Papilionidae) are one of the largest families of Lepidoptera with 572 described species recorded around the world (Scriber, 1995; Parsons, 1999). Among many tropical countries with high biodiversity, Indonesia is known as the country with both the richest fauna (121 species) and the highest species endemism (53 species) of swallowtail butterflies in the world (Collins & Smith, 1995). In spite of such rich fauna of swallowtails, studies on their biology and ecology are still limited in Indonesia except for sporadic information on host plants and immature stages for some species (Straatman & Nieuwenhuis, 1961; Straatman, 1968; Parsons, 1995b; Salmah et al., 2002; Matsuka, 2001).

Information on the duration of the immature stages is basic but indispensable for comparative studies of life histories, population dynamics, and evolutionary biology between tropical and temperate species. However, information on the duration of the immature stages of Papilionidae is still relatively inadequate in both temperate and tropical species, which seems to be caused by (1) the insertion of long pupal diapause ranging from several months (Fukuda et al., 1982) to a few years (Nakamura & Ae, 1977); (2) multivoltinism including both diapause and non-diapause generations

(cf. Fukuda et al., 1982; Masaki & Yata, 1988); or (3) facultative pupal diapause controlled by various environmental cues such as photoperiod, temperature, rainfall, nutritional, and airborne chemicals (cf. Denlinger, 1986). As a result, many studies on the developmental times of Papilionidae lack the data of pupal stages in spite of the exact description of egg and larval periods (cf. Parsons, 1999; Fukuda et al., 1982). In addition, information on tropical species is generally scantier than that of temperate counterparts. Life history information of butterflies is important not only for conservation planning but also for the promotion of ecotourism and ecoproduction including sales of butterfly specimens of endangered species reared by local people (Parsons, 1995a, 1999).

Following the first study on host plants and morphology of the immature stages for 10 Sumatran Papilionidae species (Salmah et al., 2002), this study reports the durations of the immature stages of 11 Papilionidae including two protected species (*Trogonoptera brookiana* and *Troides amphrysus*) with discussion of an extra instar and diapause.

MATERIALS AND METHODS

Study species: We studied the duration of the immature stages of eleven swallowtail species including seven Papilionini species (*Papilio demoleus, P. demolion, P. karna, P. memnon*, *P. nephelus, P. palinurus,* and *P. polytes*)*,* one Graphiini species (*Graphium agamemnon),* and three Troidini species *(Pachliopta aristolochiae, Trogonoptera brookiana* and *Troides amphrysus*).

Rearing of adults in a field cage: Four adult females of *P. demoleus* and two *P. nephelus* were caught in the field near the campus of Andalas University, Padang, West Sumatra (250 m asl.) and were released into the field cage (12 x 7.5 x 5.3 m) with eight *Citrus aurantifolia* (Rutaceae) trees inside. In order not to confuse the eggs of the two species, they were reared in the different periods in 2002.

Egg collection in the field: The eggs of nine species were collected as listed below: *P. demolion* and *P. polytes* near the campus of Andalas University, Padang (250-340 m asl.); *G. agamemnon* and *P. memnon* in Padang (40-50 m asl.); *P. palinurus* in Batu Sangkar (about 100 km north of Padang, ca. 500 m asl.) and Inderapura (about 185 km south of Padang, ca. 25 m asl.); and *P. karna, P. aristo-lochiae, T. brookiana,* and *T. amphrysus* in Harau (about 140 km north of Padang, ca. 500 m asl.).

Rearing conditions: The eggs collected in Padang both from the adult rearing cage and from the field were reared in the laboratory in Andalas Univeristy, and those collected in Harau, Batu Sangkar, and Inderapura were housed near the collection sites (Table 1). In all locations, eggs were placed in plastic cases (14 x 10 x 4.5 cm) and newly hatched larvae were kept individually in these cases with the same food plant species from which the eggs were collected (Table 1). Third, fourth, and fifth instar larvae of *T. brookiana* and *T. amphrysus* were reared in bigger plastic cases (22 x 16 x 7 cm). Food plants were exchanged every day. Fully matured final instar larvae (fifth or sixth instar, see below) of all species were moved to cages (75 x 75 x 100 cm covered with nylon net) with the leaves of their host plants.

Table 1
Butterflies species, host plant species used for rearing larvae, and rearing conditions

Species	Food plant species for rearing*	Rearing period	Location	Temperature (^{0}C)
Tribe Papilionini				
Papilio demoleus	*Citrus aurantifolia* (1)	Jul. - Aug. 2002	Padang	26-28
P. demolion	*Evodia malayana* (1)	Jun. - Jul. 1998	Padang	24-28
P. karna	*Evodia malayana* (1)	Feb. - Apr. 1998	Harau	22-27
P. memnon	*Citrus aurantifolia* (1)	Aug. - Oct. 1999	Padang	24-28
P. nephelus	*Citrus aurantifolia* (1)	Jun. - Dec. 2002	Padang	26-28
P. palinurus	*Clausena excavata* (1)	May - Jul. 2003	Batu Sangkar	21-31
P. palinurus	*Clausena excavata* (1)	Jun. – Jul. 2006	Inderapura	24-35
P. polytes	*Citrus sinensis* (1)	May -Jun. 2000	Padang	24-28
P. polytes	*Clausena excavata* (1)	May -Jun. 2000	Padang	24-28
Tribe Graphiini				
Graphium agamemnon	*Annona muricata* (2)	Jul. - Aug. 1993	Padang	26-28
Tribe Troidini				
Pachliopta aristolochiae	*Apama corymbosa* (3)	Jul. - Sept. 1999	Harau	21-24
Trogonoptera brookiana	*Aristolochia foveolata* (3)	Aug. - Oct. 2000	Harau	24-30
Troides amphrysus	*Aristolochia foveolata* (3)	Jul. - Sept. 2000	Harau	23-30

* Plant families: 1 – Rutaceae; 2 – Annonaceae; 3 – Aristolochiaceae.

Dates of hatching, larval molt, and adult emergence were recorded daily to determine the developmental times. Since some individuals died because of parasitism, disease, or unknown factors during rearing, only individuals reaching adulthood were used for the analysis. Table 1 presents the rearing periods, host plant used, and rearing conditions. All rearings were carried out under uncontrolled room temperature (ranging from 24 to 28°C in Padang, 21-30°C in Harau, 21-31°C in Batu Sangkar, and 24-35°C in Inderapura) and natural day length. It should be noted that *P. palinurus* was reared using the same food plants at two locations (Batu Sangkar, ca. 500 m asl.) and Inderapura (ca. 25 m asl.) with different temperature conditions due to the difference in elevation, and *P. polytes* was raised on two different food plants under the same rearing conditions at one location (Padang).

RESULTS

Table 2 summarizes number of instars and the duration of total developmental times (from egg to adult emergence) of 11 species. Ten out of 11 species had five instars, while all individuals of *P. aristolochiae* went through six instars.

Average egg incubation periods of the seven *Papilio* species ranged from 3.0 days (*P. demoleus*) to 7.0 days (*P. demolion*), while that of *G. agamemnon* was 4.4 day, *P. aristolochiae* 5.6 days, *T. brookiana* 7.0 days, and *T. amphrysus* 8.0 days. The egg period of *P. palinurus* reared in Batu Sangkar was 2.4 days longer than that in Inderapura (Table 2). The duration of egg incubation of *P. polytes* fed on the two different food plants was the same (4.0 days).

Table 2

Duration (Average ± SD) of successive immature stages of 11 Sumatran swallowtail butterfly species

Species	Duration of immature stages (days)											Sex ratio	
	n	Egg	L1	L2	L3	L4	L5	L6	Larval period	Pupa	Total (range)	M	F
Papilionini													
Papilio demoleus	20	3.0 ± 0.0	2.3 ± 0.5	2.3 ± 0.7	2.6 ± 0.4	2.0 ± 0.5	4.7 ± 0.7	–	13.5 ± 1.4	12.2 ± 0.4	29.7 ±1.4 (28-32)	10	10
P. demolion	4	7.0 ± 0.0	4.8 ± 0.5	3.5 ± 0.6	3.3 ±1.0	4.0 ± 0.8	4.5 ± 0.6	–	20.0 ±0.8	15.3 ± 1.0	43.0 ± 1.8 (41-45)	3	1
P. karna	15	5.2 ± 0.4	3.8 ± 0.6	4.7 ± 0.7	5.3 ± 1.0	6.1 ± 0.7	9.1 ± 0.7	–	28.9 ± 1.0	14.3 ± 0.5	49.4 ± 1.7 (48-53)	11	4
P. memnon	16	5.0 ± 0.0	4.0 ± 0.6	2.6 ± 0.6	3.3 ± 0.8	4.6 ± 0.8	6.8 ± 1.3	–	21.3 ± 2.2	14.1 ± 0.7	41.4 ± 2.2 (37-47)	8	8
P. nephelus	16	4.0 ± 0.0	2.6 ± 0.5	2.6 ± 0.5	2.7 ± 0.5	4.4 ± 0.6	6.8 ±1.2	–	19.1 ± 2.3	53.9 ± 38.0	78.0 ± 37.8 (35-141)	8	8
P. palinurus	5*	6.2 ± 0.4	4.6 ± 0.6	4.2 ± 0.8	4.4 ± 0.6	5.2 ± 1.3	5.6 ± 0.6	–	24.0 ± 2.4	13.8 ± 0.8	45.0 ± 1.6 (43-47)	3	2
P. palinurus	31**	3.8 ± 0.4	3.5 ± 0.5	3.8 ± 0.5	3.7 ± 0.6	4.0 ± 0.6	6.8 ± 1.0	–	21.7 ± 2.3	12.7 ± 1.0	39.2 ± 2.5 (34-43)	17	14
P. polytes	7***	4.0 ± 0.0	2.9 ± 0.4	3.0 ± 0.0	2.7 ± 0.5	2.7 ± 0.5	6.3 ± 0.8	–	17.6 ± 0.8	12.0 ± 0.6	34.6 ± 1.1 (33-36)	5	2
P. polytes	8****	4.0 ± 0.0	2.5 ± 0.5	2.5 ± 0.5	2.3 ± 0.5	2.8 ± 0.7	5.6 ± 1.1	–	15.6 ± 1.6	12.4 ± 0.7	32.0 ± 1.9 (30-35)	4	4
Graphiini													
Graphium agamemnon	5	4.4 ± 0.6	2.8 ± 0.8	3.6 ± 0.9	2.8 ± 0.5	4.2 ± 0.8	9.2 ± 1.6		22.6 ± 2.1	11.6 ± 0.6	39.8 ± 2.5 (38-44)	3	2
Troidini													
Pachliopta aristolochiae	15	5.6 ± 0.8	4.5 ± 0.6	4.8 ±_0.7	4.4 ± 0.6	5.8 ± 0.8	6.3 ± 1.2	9.1 ± 1.5	34.9 ± 2.7	17.0 ± 0.8	59.5 ± 3.0 (54-62)	9	6
Trogonoptera brookiana	9	7 .0 ± 0.0	2.9 ± 0.8	3.8 ± 0.4	4.3 ± 0.5	5.0 ± 0.5	7.1 ± 0.3	–	23.7 ± 1.2	23.7 ± 1.0	56.3 ± 2.1 (54-61)	5	4
Troides amphrysus	3	8.0 ± 0.0	4.0 ±1.0	4.3 ± 0.6	5.0 ± 1.0	8.7 ± 0.6	10.3 ± 0.6	–	32.3 ± 2.25	28.0 ± 1.0	70.3 ± 2.1 (68-72)	2	1

Reared at Batu Sangkar* and Inderapura**, reared on *Clausena excavata* *** and *Citrus sinensis* ****

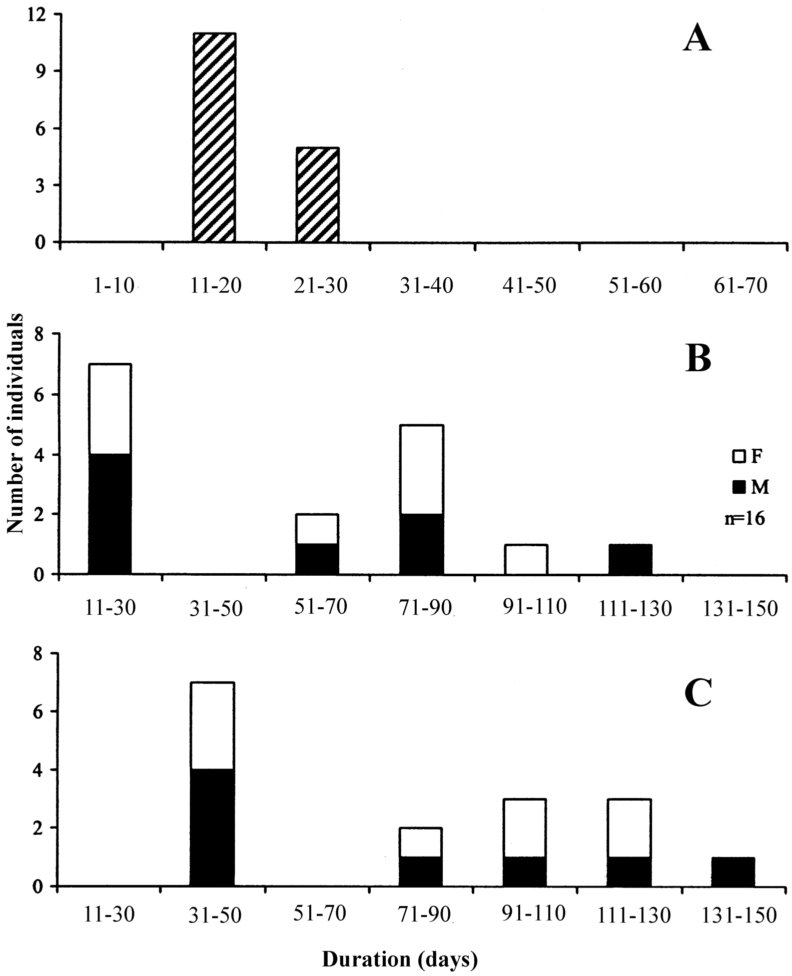


Fig. 1. Variation in the duration of larval period (A), pupal period (B), and whole immature stage (C) of *Papilio nephelus* reared on *Clausena excavata*. F, M – female and male, respectively; n – number of individuals reared.

The average duration of the larval stages in the seven *Papilio* species ranged from 13.5 days (*P. demoleus*) to 28.9 days (*P. karna*), and that of *G. agamemnon* was 22.6 days. Those of Troidini species were longer than those of some Papilionini and Graphiini species, i.e. *T. brookiana* 23.7 days, *T. amphrysus* 32.3 days, and *P. aristolochiae* 34.9 days. The average duration of larval stages of *P. palinurus* reared at the two locations (Batu Sangkar and Inderapura, Table 1) differed by 2.3 days (Table 2) and those of *P. polytes* reared using two different food plants (Table 1) differed by 2.0 days (Table 2).

The average pupal periods of the seven *Papilio* species ranged from 12.0 days (*P. polytes*) to 53.9 days (*P. nephelus*). Those of *P. palinurus* differed by 1.1 days between the two locations. The frequency distribution of pupal duration for *P. nephelus* was bimodal with peaks at 11-30 and 71-90 days, which generated a large SD value (38.0 days) (Fig. 1B). This large variation occurred in the pupal stage because frequency distribution of the larval stage was clearly unimodal at 19.1±2.3 days (average±SD) (Fig. 1A). The individuals whose pupal duration was more than 25 days were considered to enter diapause (see Discussion). The second longest was *P. demolion* (15.3 days). The pupal durations of Troidini species were; *P. aristolochiae,* 17.0 days; *T. brookiana,* 23.7 days; and *T. amphrysus,* 28.0 days.

Table 2 summarizes the average total developmental times (from egg to adult emergence) of 11 species: Papilionini species, 43.6 days; Graphiini, 39.8 days; and Troidini, 59.5-70.3 days; and *P. palinurus,* 45.0 days (Batu Sangkar) and 39.2 days (Inderapura). The frequency distribution of total developmental times of *P. nephelus* was bimodal at 31-50 days and flat peaks at 91-110 and 111-130 days (average and SD: 78.0±37.8) (Fig. 1C). This distribution was brought about after the pupal stage (Fig. 1B), because the frequency distribution of the larval period was narrowly unimodal (Fig. 1A).

DISCUSSION

The present study clearly shows that the immature durations of the Papilionini and Graphiini are much shorter than those of the Troidini (Table 2). Matsuka (2001) mentioned that the duration of immature stages in birdwing butterflies (*Ornithoptera*, *Trogonoptera*, and *Troides*) were usually in the ranges; egg, 7-12 days; larva, 30-40 days; and pupa, 30-45 days with a few exceptions such as *Ornithoptera alexandrae* (70-74 days in larval stage, Straatman 1971; Parsons, 1984) and *Troides vandepolli* (1-8 months in pupal stage, Arbaimun pers. com. after Matsuka, 2001).

The present study reported that 10 study species had five instars, but only one species, *P. aristolochiae,* had six instars (Table 2). An Indian *P. aristolochiae* population reared on *Aristolochia indica* (Aristolochiaceae) was reported to have 5 instars (Atluri et al., 2001). Since all individuals of *P. aristolochiae* that were fed on *Apama corymbosa* completed the immature stages normally and adults included both sexes (9 males and 6 females), *P. aristolochiae* in this study is considered to be a population with six larval instars, which confirmed intraspecific variability in the number of instars in this species.

Diapause in the tropics has been reported frequently, although it is less common compared to regions at higher latitude (Denlinger, 1986). Diapausing individuals are separated from non-diapausing ones by a boundary of 25 days (Yoshio & Ishii, 1998). Taking this criterion, among 16 *P. nephelus* individuals in the present study nine entered diapause (Fig. 1), and to our knowledge this is the first diapause record in Papilionidae observed at low latitude (0°40'S). This study location is near the equator so that day length variation is less than 30 minutes and there is no strong dry

season. Induction of diapause in tropical butterflies was explained by several factors such as short day length coupled with low temperature (Singh, 1993), low temperature (Ishii, 1987), and dry season (Blau, 1980). The factors inducing diapause in a part of *P. nephelus* population are to be clarified

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REFERENCES

Atluri, J.B., Venkata Ramana, S.P. & Subba Reddi, C. 2001. Life history of *Pachliopta aristolochiae* (Lepidoptera: Rhopalocera: Papilionidae) from India. – Journal of the National Taiwan Museum 54: 9-12.

Blau, W.S. 1980. Notes on the natural history of *Papilio polyxenes stabilis* (Papilionidae) in Costa Rica. – Journal of the Lepidopterists' Society 34: 321-324.

Collins, N.M. & Smith, H.M. 1995. Threats and priorities in conserving swallowtails. – In: Scriber, J.M., Tsubaki, Y. & Lederhouse, R.C. (Eds.). Swallowtail Butterflies: Their Ecology and Evolutionary Biology. Scientific Publishers: Gainesville, Washington, Hamburg, Lima, Taipei, Tokyo: 345-357.

Denlinger, D.L. 1986. Dormancy in tropical insects. – Annual Review of Entomology 31: 239-264.

Fukuda, H., Hama, E., Kazuya, T., Takahashi, A., Takahashi, M., Tanaka, B., Tanaka, H., Wakabayashi, M. & Watanabe, Y. 1982. The Life Histories of Butterflies in Japan. Vol. 1. Hoikusha Publisher, Osaka. (In Japanese).

Ishii, M. 1987. Diapause potential in tropical papilionids (Lepidoptera: Papilionidae). – Applied Entomology and Zoology 22: 114-115.

Masaki, S. & Yata, O. 1988. Seasonal adaptation and photoperiodism in butterflies. – Special Bulletin of Lepidopterological Society of Japan 6: 341-383. (In Japanese).

Matsuka, H. 2001. Natural History of Birdwing Butterflies. Matsuka Publisher, Tokyo. (In Japanese).

Nakamura, I. & Ae, S.A. 1977. Prolonged pupal diapause of *Papilio alexanor*: Arid zone adaptation directed by larval host plant. – Annals of the Entomological Society of America 70: 481-484.

Parsons, M.J. 1984. The biology and conservation of *Ornithoptera alexandrae*. – In: Vane-Wright, R.I. & Ackery, P.R. (Eds.). The Biology of Butterflies. Symposium of the Royal Entomological Society of London, No. 11. Academic Press, London: 327-331.

Parsons, M.J. 1995a. Butterfly farming and trading in the Indo-Australian region and its benefits in the conservation of swallowtails and their tropical forest habitats. – In: Scriber, J.M., Tsubaki, Y. & Lederhouse, R.C. (Eds.). Swallowtail Butterflies: Their Ecology and Evolutionary Biology. Scientific Publishers: Gainesville, Washington, Hamburg, Lima, Taipei, Tokyo: 371-391.

Parsons, M.J. 1995b. The early stages and ecology of *Ornithoptera tithonus*. (pp.). – In: Scriber, J.M., Tsubaki, Y. & Lederhouse, R.C. (Eds.). Swallowtail Butterflies: Their Ecology and Evolutionary Biology. Scientific Publishers: Gainesville, Washington, Hamburg, Lima, Taipei, Tokyo: 401-404.

Parsons, M.J. 1999. The Butterflies of Papua New Guinea: Their Systematics and Biology. Academic Press, London.

Salmah, S., Abbas, I. & Dahelmi. 2002. Butterflies in Kerinci Seblat National Park. Yayasan Kehati, Jakarta. (In Indonesian).

Scriber, J.M. 1995. Overview of swallowtail butterflies: Taxonomic and distributional latitude. – In: Scriber, J.M., Tsubaki, Y. & Lederhouse, R.C. (Eds.). Swallowtail Butterflies: Their Ecology and Evolutionary Biology. Scientific Publishers: Gainesville, Washington, Hamburg, Lima, Taipei, Tokyo: 1-8.

Singh, S.P. 1993. Species composition and diapause in citrus butterflies. – Journal of Insect Science 6: 48-52.

Straatman, R. 1968. On the biology of some species of Papilionidae from the island of Celebes (East-Indonesia). – Entomologische Berichten 28: 229-233.

Straatman, R. 1971. The life history of *Ornithoptera alexandrae* Rothschild. – Journal of the Lepidopterists' Society 25: 58-64.

Straatman, R. & Nieuwenhuis, E. J. 1961. Biology of certain Sumatran species of *Atrophaneura*, *Trogonoptera* and *Troides* (Lepidoptera, Papilionidae). – Tijdschrift voor Entomologie 104: 31-41.

Yoshio, M. & Ishii, M. 1998. Geographical variation of pupal diapause in the great mormon butterfly, *Papilio memnon* L. (Lepidoptera: Papilionidae), in Western Japan. – Applied Entomology and Zoology 33: 281-288.